# ORIGINAL PAPER

# Biomass accumulation and nutrient uptake of 16 riparian woody plant species in Northeast China

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Abstract: Our research focused on eutrophication control and species screening for riparian zone vegetation restoration in the upstream reach of the Hun River. We studied 16 hardwood plant species to investigate nutrient concentrations and nitrogen and phosphorus accumulations. After about 120 days of growth in pots, these 16 species varied in dry matter biomass, ranging from 15.13 to 637.16 g. Total nitrogen (TN) and total phosphorus (TP) concentrations and distribution in roots, stems and foliage differed both within and between tested species. Mean TN and TP accumulation ranged from 0.167 to 14.730 g per plant and from 0.016 to 1.20 g, respectively. All 16 species, but especially *Lespedeza bicolor*, *Robinia pseudoacacia* and *Sorbaria sorbifolia* had strong potential to remove TN and TP from soil and could be widely utilized for the restoration of destroyed riparian zones in northeast China.

Keywords: Nitrogen, phosphorous, ecological restoration, foliage

### Introduction

Eutrophication is a widespread and increasing problem in water resources in many countries (Bennett et al. 2001). Agricultural non-point source pollution remains the greatest global contributor to eutrophication (Corwin et al. 1997; Dabrowski et al. 2002).

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The riparian zone receives and retains large amounts of nutrient inputs from farmland. These inputs can indirectly lead to eutrophication of rivers (Giese et al. 2003; McClain et al. 2003). Riparian zone vegetation, through interception and storage, plays an important role in protecting the river water from eutrophication (Lowrance et al. 1997; Hazlett et al. 2008; McBroom et al. 2008). Nitrogen concentration in runoff water can be reduced by 65%-100% after passing through a riparian zone forest (Spoelstra et al. 2010). Depending on the plant species, plant nutrient uptake has been shown to account for 3%-47% of nitrogen removal and 3%-60% of phosphorus removal from runoff water in the riparian zone (Cooke 1992; Tanner 1996; Kuusemets et al. 2001). The riparian zone forms an appropriate environment for nutrient removal (Lowrance et al. 1984; Lee et al. 2009). Previous studies have examined more than 50 aquatic plants (Borin and Salvato 2012) and herbs (Mcjannet et al. 1995; Yu et al. 2014). On an annual basis, forested riparian buffer strips have proven more effective at reducing nitrogen concentrations in streams than herbaceous buffers (Lowrance et al. 1984; Osborne and Kovacic 1993; Hefting et al. 2005). Poplar forest in the riparian zone proved more effective (99% retention of NO<sub>3</sub><sup>-</sup>) than grass (84% retention of NO<sub>3</sub><sup>-</sup>) during winter months (Haycock and Pinay 1993).

Selecting suitable woody plants for restoration of polluted, damaged riparian zones is an effective and efficient measure for controlling non-point-source agricultural inputs of nutrients (Bedford et al. 1999). However, few studies directly compared the effect of various plant species under similar conditions for extended periods of time to identify those which most efficiently accumulate nitrogen and phosphorus. This is especially true in cold temperate regions.

To fill the above information gap, we studied 16 native woody plants of 9 families to assess their performance in removing nitrogen and phosphorus from runoff water. We quantified Total nitrogen and total phosphorus concentrations, and biomass accumulations of the plants in different tissues. This research was undertaken to aid in predicting the response of species to eutrophication and providing statistical support for plant species screening for the restoration of eutrophicated riparian zones.



#### Materials and methods

#### Plant culture

Based on species lists for riparian zone forests along the Hun River, we selected sixteen woody plant species of northeastern China for study: Syringa reticulate, Prunus padus, Robinia pseudoacacia, Pterocarya stenoptera, Juglans mandshuriea, Berberis dielsiana, Sambucus williamsii, Salix matsudana, Quercus mongolica, Rosa davurica, Euonymus alatus, Acer truncatum, Populus alba, Lespedeza bicolor, Ulmus pumila, and Sorbaria sorbifolia. The uniform plants (three-year-old seedlings) used in this study were bought from a nursery and cultivated in pots (five pots per species) with 8-kg un-contaminated soil (Meadow burozem soil, 6.82 pH, 2.53% organic carbon, 4.35% organic matter, 1.88 mg·g<sup>-1</sup> total nitrogen, 0.24 mg·g<sup>-1</sup> total phosphorus. After one month of growth in pots, a screening experiment was conducted from May to October 2012 at Shenyang Arboretums of Chinese Academy of Sciences (41°54' N, 123°35' E). Plants were grown under nutrient controlled conditions with nutrient solution added.

Nutrient solution was used to simulate the high nutrient input levels which typically occurred in agriculture runoff. The standard solution contained 56 mg·L<sup>-1</sup> of nitrogen (as NH<sub>4</sub>NO<sub>3</sub>) and 62 mg·L<sup>-1</sup> of phosphorus (as NaH<sub>2</sub>PO<sub>4</sub>·2H<sub>2</sub>O), a control group was set up (CK: 0 mg·L<sup>-1</sup> of nitrogen and 0 mg·L<sup>-1</sup> of phosphorus). The nutrient addition was applied once a week. The experiment was replicated three times.

# Nutrient analysis

At the end of the growing season (October), the plant samples were harvested, washed and separated into roots, stem and foliage, heated in an oven at 90°C for 30 min and dried at 65°C to constant weight. Oven-dried materials were milled and passed through a 100-mesh (0.149 mm) nylon sieve (Huafeng, Zhejiang, China), and then stored in jars prior to laboratory analyses (Lu 1999).

Subsamples were digested for total nitrogen (TN) and total phosphorus (TP) measurement according to the sulfuric acid-hydrogen peroxide (H<sub>2</sub>SO<sub>4</sub>-H<sub>2</sub>O<sub>2</sub>) method (Son and Gower 1992). TN was quantified using the semi-micro Macro Kjeldahl method (Ruizheng Kjeldahl nitrogen analyzer KDY-600D, Shanghai, China). TP was quantified using the molybdenum antimony-ascorbic acid colorimetric method (MADAC) (SHI-MADZU UV-1800 spectrophotometer, Japan).

#### Statistical analyses

Average values and standard errors (S.E.) were calculated by Microsoft Office Excel 2007 for all data. Statistical procedures used in this study were performed using SPSS (Version 16.0, SPSS Inc. 2007). Standard one-way analyses of variance (ANOVA) were used to test significance of differences among



and between the sixteen tested species and between roots, stems and foliage, with respect to TN, TP and biomass. Duncan multiple range test was employed to show the variation in TN and TP between species. Spearman's correlation analysis was used to determine the correlationships between tissues and nutrition. Significant and extremely significant differences were set as p < 0.05 and p < 0.01, respectively. Hierarchical cluster analysis was used to classify plants into different groups based on the nutrient distribution in tissues. Origin 8.0 was used to draw figures.

#### Results

#### Total biomass

At the end of the research, the total biomass per plant was compared between the 16 tested species (Fig. 1). After about 120 days of growth, total biomass/plant differed significantly by species and treatment (p < 0.05, n=3, Fig.1). The biomass of CK ranged from 4.20 to 237.89 g in the order: Quercus mongolica < Ulmus pumila <Pterocarya stenoptera <Sambucus williamsii <Euonymus alatus < Berberis dielsiana < Prunus padus < Rosa davurica <Acer truncatum <Juglans mandshuriea < Syringa reticulate <Salix matsudana <Populus alba <Sorbaria sorbifolia < Robinia pseudoacacia < Lespedeza bicolor. The T1 (treatment) biomass per plant value ranged from 15.13 to 637.16 g in the following order: Pterocarya stenoptera < Quercus mongolica <Ulmus pumila <Berberis dielsiana <Sambucus williamsii < Prunus padus <Rosa davurica < Euonymus alatus < Acer truncatum<Juglans <mandshuriea < Syringa reticulate <Salix matsudana < Populus alba <Sorbaria sorbifolia <Lespedeza bicolor < Robinia pseudoacacia. T1 treatment resulted in greater biomass than CK. Robinia pseudoacacia and Lespedeza bicolor vielded more biomass than other species.

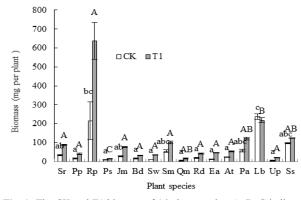


Fig. 1: The CK and T1 biomass of 16 plant species. A, B, C indicate T1 the significance different at p < 0.05; a, b, c indicate CK the significance different at p < 0.05. Error bars denote 1 SE (n=3). Syringa reticulate (Sr), Prunus padus (Pp), Robinia pseudoacacia (Rp), Pterocarya stenoptera (Ps), Juglans mandshuriea (Jm), Berberis dielsiana (Bd), Sambucus williamsii, Salix matsudana (Sm), Quercus mongolica (Qm), Rosa davurica (Rd), Euonymus alatus (Ea), Acer truncatum (At), Populus alba (Pa), Lespedeza bicolor (Lb), Ulmus pumila (Up) and Sorbaria sorbifolia (Ss).

## Variation in TN and TP concentration among tissues

For all tested species, TN and TP concentrations varied between roots, stem and foliage (Fig. 2). TN (p <0.05) and TP (p <0.05) concentrations in foliage were significantly greater than in roots and stems. There was no consistent pattern in the distribution of N or P concentrations in stems and roots. The plant tissues in T1 treatment had higher N and P concentrations than that did CK. The foliage of *Sambucus williamsii* and *Berberis dielsiana* had the highest TN (35.56 mg·g<sup>-1</sup>) and TP (5.49 mg·g<sup>-1</sup>) concentrations. The average TN concentration in roots, stems and foliage was 12.11 mg·g<sup>-1</sup>, 12.96 mg·g<sup>-1</sup> and 22.46 mg·g<sup>-1</sup>; The TP concentration in roots, stems and foliage was 1.58 mg·g<sup>-1</sup>, 1.51 mg·g<sup>-1</sup> and 2.51 mg·g<sup>-1</sup>, respectively.

# Accumulation of TN and TP in plants

According to the analysis of TN and TP accumulations in the plants, there were significant differences in the roots, stem, foliage and among species. As shown in Tables 1 and 2, mean TN accumulated in the whole plant ranged from 166.65 to 14729.73 mg (Quercus mongolica <Pterocarya stenoptera <Ulmus pumila< Rosa davurica < Prunus padus < Sambucus williamsii < Berberis dielsiana <Acer truncatum <Euonymus alatus <Juglans mandshuriea <Syringa reticulate <Salix matsudana <Populus alba <Sorbaria sorbifolia < Lespedeza bicolor <</p> Robinia pseudoacacia) and TP ranged from 16.06 to 1203.54 mg (Quercus mongolica <Pterocarya stenoptera pumila < Euonymus alatus < Rosa davurica < Sambucus williamsii < Prunus padus <Berberis dielsiana <Acer truncatum <Salix matsudana < Populus alba < Juglans mandshuriea < Syringa reticulate <Sorbaria sorbifolia <Lespedeza bicolor <Robinia pseudoacacia). Species Rp showed by a wide margin the highest accumulated TN and TP.

Spearman correlation analysis indicated that TN and TP accumulations in different tissues were positively correlated (Table 3). The Correlation coefficient ranged from 0.81 to 0.96.

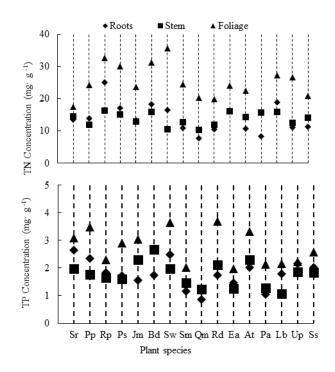


Fig. 2: Total Nitrogen and Total Phosphorus concentration (dry, wt) of plants. Syringa reticulate (Sr), Prunus padus (Pp), Robinia pseudoacacia (Rp), Pterocarya stenoptera (Ps), Juglans mandshuriea (Jm), Berberis dielsiana (Bd), Sambucus williamsii, Salix matsudana (Sm), Quercus mongolica (Qm), Rosa davurica (Rd), Euonymus alatus (Ea), Acer truncatum (At), Populus alba (Pa), Lespedeza bicolor (Lb), Ulmus pumila (Up) and Sorbaria sorbifolia (Ss).

Table 1: Comparison of the TN accumulations (mg) in different tissues (roots, stem and foliage) of the sixteen tested tree species (means ± sd)

Total Nitrogen	Roots (mg)	Stem (mg)	Foliage (mg)	Total (mg)
Syringa reticulata	607.67±46.65c	438.08±6.93b	230.54±3.47a	1276.29±50.12A
Prunus padus	227.91±20.69b	205.41±0.17ab	149.19±16.92a	582.52±37.43A
Robinia pseudoacacia	3898.98±546.31a	4861.19±1151a	5969.57±1169.87a	14729.73±1926.56C
Pterocarya stenoptera	141.77±6.18a	65.22±16.24a	74.24±28.32a	281.22±38.39A
Juglans mandshuriea	508.55±48.41b	245.39±20.33a	461.4±45.14b	1215.34±23.59A
Berberis dielsiana	256.8±11.13b	179.4±18.89a	209.61±18.77ab	645.81±48.79A
Sambucus williamsii	293.58±4.6c	112.75±0.38a	228.16±17.8b	634.49±13.58A
Salix matsudana	590.11±209.36a	337.42±54.43a	418.65±182.68a	1346.19±81.11A
Quercus mongolica	68.13±29.46a	40.31±3.71a	58.22±3.35a	166.65±29.82A
Rosa davurica	238.99±14.11a	181.34±16.28a	77.14±9.21b	497.47±39.59A
Euonymus alatus	417.21±10.66b	202.32±5.72a	193.78±14.47a	813.31±1.91A
Acer truncatum	255.09±13.34a	167.04±9.83a	379.43±42.44b	801.56±65.6A
Populus alba	517.97±32.11b	814.6±120.35b	120.12±2.51a	1452.68±149.95A
Lespedeza bicolor	998.74±119.38a	1788.72±197.67b	1466.89±190.63ab	3956.23±358.43B
Ulmus pumila	78.68±4.14a	87.64±16.89a	176.23±25.32b	342.55±46.35A
Sorbaria sorbifolia	643.41±25.35b	704.77±38.06b	354.05±32.57a	1702.24±95.99A

**Notes**: a, b, c in the same row indicate the significance different at p < 0.05. A, B, C in the same line indicates the significance different at p < 0.05.



 Table 2: Comparison of the TP accumulations (mg) in different tissues (roots, stem and foliage) of the sixteen tested tree species (means  $\pm$  sd)

Total Phosphorus	Roots (mg)	Stem (mg)	Foliage (mg)	Total (mg)
Syringa reticulata	120.28±9.1b	59.24±0.35a	40.77±3.14a	220.16±5.61AB
Prunus padus	38.34±4.44b	30.69±0.72ab	21.32±2.61a	90.2±6.33AB
Robinia pseudoacacia	256.99±16.08a	502.79±123.27a	441.38±102.13a	1203.54±191.09C
Pterocarya stenoptera	9.76±0.56a	6±1.57a	8.21±2.44a	28.26±3.45A
Juglans mandshuriea	60.07±4.33a	43.73±2.82a	59.1±7.92a	162.94±0.77AB
Berberis dielsiana	24.26±1.71a	30.04±4.32a	36.73±4.75a	91.32±10.77AB
Sambucus williamsii	44.34±0.56b	21.17±1.09a	23.39±1.58a	88.91±0.07AB
Salix matsudana	41.53±0.83a	80.72±8.82b	21.55±1.33a	143.61±9.32AB
Quercus mongolica	7.27±2.87a	4.8±0.52a	3.73±0.2a	16.06±3.59A
Rosa davurica	39.69±1.54c	32.32±1.54b	14.6±0.75a	86.51±2.33AB
Euonymus alatus	37.4±1.06b	14.79±0.16a	17.09±0.61a	69.27±1.84A
Acer truncatum	48.8±2.83b	26.64±0.24a	56.56±4.31b	132.07±7.38AB
Populus alba	65.09±2.44a	65.76±7.9a	16.41±1.13b	147.17±6.59AB
Lespedeza bicolor	94.41±16.4a	119.29±21.75a	115.48±21.53a	328.39±21.13B
Ulmus pumila	15.36±0.04a	$13.08 \pm 1.87a$	14.72±2.48a	43.14±4.39A
Sorbaria sorbifolia	114.22±2.52c	91.59±2.21b	43.59±4.14a	249.35±3.84AB

**Notes**: a, b, c in the same row indicate the significance different at p < 0.05. A, B, C in the same line indicates the significance different at p < 0.05.

#### Distribution of TN and TP in tissues

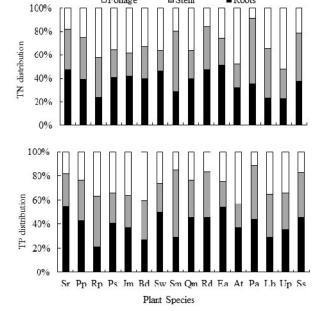
The accumulated quantities of TN in roots, stems and foliage accounted for 23–48%, 18–56% and 8–52%, respectively, of the total accumulations (Fig. 3). The proportions of TP in roots, stems, and foliage were 21–55%, 20–56% and 11–43%, respectively, of the total accumulations (Fig. 3).

**Table 3:** Spearman correlation coefficient of TN and TP accumulations in different tissues.

Tissues	Nutrient	Roots		Stem		Foliage	
		TN	TP	TN	TP	TN	TP
Roots	TN	1	0.92**	0.91**	0.89**	0.89**	0.88**
	TP		1	0.87**	0.90**	0.84**	0.84**
Stem	TN			1	0.95**	0.85**	0.81**
	TP				1	0.85**	0.84**
Foliage	TN					1	0.96**
	TP			_			1

Notes: \*\* Correlation is significant at the 0.01 level (2-tailed). TN is Total Nitrogen; TP is Total Phosphorus.

Based on the distribution of nutrient accumulations, the tested species were clustered into three distinct groups by Hierarchical cluster analysis (Fig. 4). The first group, including only two species, had a relatively higher proportion of nutrients (more than 50%) in stems; the second group included 9 species in which roots had the highest percentages of nutrients (about 45%). Foliage and stems shared almost the equal proportions. The third group, including 5 species, shared nearly equal percentages of nutrients in roots, stems and foliage. Species of the same families were clustered into the same groups (except *Quercus mongolica*).



□Foliage

Fig. 3: Total nitrogen and total phosphorus distribution in roots, stem and foliage. Syringa reticulate (Sr), Prunus padus (Pp), Robinia pseudoacacia (Rp), Pterocarya stenoptera (Ps), Juglans mandshuriea (Jm), Berberis dielsiana (Bd), Sambucus williamsii, Salix matsudana (Sm), Quercus mongolica (Qm), Rosa davurica (Rd), Euonymus alatus (Ea), Acer truncatum (At), Populus alba (Pa), Lespedeza bicolor (Lb), Ulmus pumila (Up) and Sorbaria sorbifolia (Ss).

## **Discussion**

# Interspecific variation

Biomass accumulations of nutrients varied by species (Fig. 1). These differences were largely determined by their physiology (i.e., net assimilation) and morphology (Ma et al. 2010), as many species have various mechanisms for adaptation, including ad-



justments of growth rate, modifications of plant structure (Li et al. 2007). Plants with high biomass accumulate more nutrients in their tissues (Jiang et al. 2011). *Robinia pseudoacacia* and *Lespedeza bicolor* yielded the highest biomass and accumulated the highest quantities of nutritients. Kyambadde et al. (2004) and Iamchaturapatr et al. (2007) report that species like *Robinia pseudoacacia* and *Lespedeza bicolor* are more suitable for riparian zone restoration owing to higher N and P removal from water.

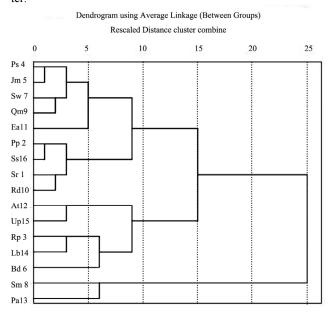


Fig. 4: Euclidean distance clustering tree. Syringa reticulate (Sr), Prunus padus (Pp), Robinia pseudoacacia (Rp), Pterocarya stenoptera (Ps), Juglans mandshuriea (Jm), Berberis dielsiana (Bd), Sambucus williamsii, Salix matsudana (Sm), Quercus mongolica (Qm), Rosa davurica (Rd), Euonymus alatus (Ea), Acer truncatum (At), Populus alba (Pa), Lespedeza bicolor (Lb), Ulmus pumila (Up), and Sorbaria sorbifolia (Ss).

Boyd (1970, 1978) observed large interspecific variation in nutrient concentrations in aquatic plants. In his study, it is unclear whether such differences were related to environmental nutrient levels or to the different absorption rates of the various species because the plants were collected from the field. However, in our study, plants were of the same age and were cultivated in the same environment conditions. In the absence of other factors such as disturbance, interspecific variation in tissue TN and TP was recorded in our study. This result was in accordance with research on 41 wetland plants reported by McJannet (1995).

In our study, each species showed dramatically different TN and TP concentrations and accumulations between tissues (roots, stems and foliage). Nutritients were recorded at higher concentrations in foliage than in roots and stems. There were, however, no consistent differences in nutrient concentrations between roots and stems. This result was consistent with the results of Zhu et al. (2011). Li et al. (2013) reported nutrition distribution as foliage> stems> roots in 30 common plant species grown in the hydro-fluctuation belt of Baihua Reservoir in Guizhou prov-

ince, China. This was because both the structure and function differ by tissue type. Leaves contain photosynthetic tissues whose metabolism is active, while roots and stems are storage tissues that transport water and nutrients. Stems and roots, which were primarily composed of cellulose, have lower nutrition demand (Shan et al. 2011).

We recorded positive correlation between TN and TP. Niinemets and Kull (2003), however, found no correlation between TN and TP in plant species in a wooded meadow and a bog, probably because concentrations were similar in all species. In fact, both N and P can stimulate growth or other processes because TN supply affects how efficiently TP is acquired and used, and vice versa (Treseder and Vitousek 2001; Gusewell et al. 2003).

The function of riparian zone plants in nutrition removal

TN and TP concentration of the 16 woody plants ranged from 8.38 to 35.56 mg·g<sup>-1</sup> and from 0.87 to 5.49 mg·g<sup>-1</sup>, respectively. TN and TP were accumulated to quantities ranging from 111.6 to 14729.73 mg and 16.06 to 1203.54 mg, respectively. Of the 16 woody plants, Lespedeza bicolor, Robinia pseudoacacia, and Sorbaria sorbifolia had absorbed most TN and TP from soil and stored most in tissues. These three species are recommended as preferred restoration plants for the main purpose of TN and TP removal in the riparian zone. Of course, the above results showed only the absorption and storage ability of the tested plant species. In other words, the data in Table 2 and Table 3 were just a part of the total removal effect by the whole riparian ecosystem. The riparian plant community provides a suitable environment for TN and TP removal (Wu et al. 2011). Other mechanisms, such as rhizosphere microbial activity and physical processes, could also contribute to the removal of most pollutants (Brix 1987; Gottschall et al. 2007). Consequently, the nutrient treatment and removal capacities of woody plants examined this study would, if grown in the wild, undoubtedly far exceed the results presented here. Due to the complexity of riparian ecosystems, more research is needed to learn more about the processes and mechanisms in natural situations.

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